Art. XV.—*On the Structure of Hologlœa dubia, an Organism of doubtful affinity.*

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(With Plates XXIV., XV.).

[Read 10th September, 1908.]

In February, 1905, Dr. T. S. Hall collected at Lorne, on the shores of the southern coast of Victoria, a few specimens of a small organism that had evidently been thrown up during heavy weather in Bass Strait. From that time to this no further specimens have been obtained. Dr. Hall was unable to preserve many, and the fourteen that he did secure were preserved in formalin, and remained unnoticed until recently in the store collections of the Biological Laboratory in the Melbourne University.

The general appearance of the organism suggested at first glance an alliance with the Ctenophora, but what appears on superficial examination to be of the nature of ctenophoral bands turn out, on minute examination, to have nothing whatever to do with these, and not to possess the slightest trace of ctenophoral plates or canals.

I thought at one time that the organisms might be detached parts of some larger form—specially modified individuals of some colonial animal—but careful search reveals no trace of any such separation having taken place, and I can only conclude, therefore, that they represent a stage in the life history of some form which is at present unrecognised; possibly, as will be seen later, a nurse stock.

Their structure is at once simple, definite and remarkable, and, in certain respects, quite unlike that of any organism at present described.

Each has taken the form of a mass of stiff jelly with four sides and an oral and aboral end. (Figs. 1, 2, 3, 4.) Every

margin is bounded by a very distinct band of finely punctated material that stands out clearly as a light band when the organism is viewed against a dark background. For the purpose of convenience in reference, I have numbered the lateral margins of the quadrangular mass, 1, 2, 3 and 4.

In Figure 1 is seen a general view of the organism. It is quite transparent, and by careful focussing under a low power the whole of the structure of the body can be seen. Figure 1 represents a side view, Figure 3 a view of the oral surface, and Figure 4 a diagrammatic transverse section. The dimensions of the largest specimen are as follows, but there is very little variation in size amongst them. Distance between oral and aboral surfaces, 11 mm.; width between margins 1 and 3, 9 mm.; downward projection of margin 1, 2.5 mm.

General Form.—When the animal is alive the body is probably a fairly regular, quadrangular shaped mass of stiff jelly. The mouth lies in the middle of the oral surface, and leads into a simple, wide, flask-shaped cavity occupying the centre of the jelly mass. From the central point of its distal end there arises a very small but distinct tube which runs up to, and opens on a conical projection on the aboral surface. (Fig. 2.)

The margin numbered 1 (Figs. 1 and 5) is prolonged beyond the oral surface so as to form a conical projection, the three sharply marked edges of which are serrated. The surface that lies between the margins 1 and 2 (Figs. 1, 2, 4 and 5) is indented by a deep groove which penetrates the jelly almost as far as, but not quite to, the central tubular cavity. On the aboral surface the jelly is lifted up (Fig. 2) to form a conical projection, which is cut through by the groove, into which, at its upper end, the small tube from the central cavity opens. The groove, which forms one of the most remarkable features of the organism, is a very definite structure, and extends to within a short distance of the oral surface. Except at the aboral end its lips are close together. At the oral end one lip is continued (Fig. 5) as a well-defined line of densely punctated material, precisely similar to that of the margins, marked with ten or eleven serrations. It passes down, curving gracefully on to the aboral projection already described, the groove not extending so far as its termination.

Structure of Body Wall.—One of the most striking features is the entire absence of any cellular structure on the surface of the body. When stained with Haemalum or Picro-carmine, transverse sections cut with the freezing microtome[1] showed, except along the marginal lines, only a homogeneous jelly extending from the central cavity to the surface, without a trace of a cell either in the jelly or on the external surface. Preparations were made of four specimens, with precisely the same result in each case. It is, of course, possible, but scarcely probable, that every vestige of cellular structure, supposing such to have once been present on the surface, may have been rubbed off. The otherwise very definite and in all cases similar form and appearance of the outside surface of the fourteen specimens seem to indicate that this remarkable structure, or rather absence of structure, is characteristic of the organism.

As already said, the margins are very definitely modified. Against a dark background they stand out white. In some cases they are slightly broken and discontinuous, but as a general rule they are regular. On the surface of each is a very definite cuticular layer, sharply marked by reason of the fact that it does not, or at most only slightly, take stain. On the oral projection, and also in some cases on the lower parts of the other marginal lines and around the margins of the oral surface the cuticle is distinctly serrated. (Figs. 5 and 5a.) Each serration has a core of material that slightly takes stain. On either side of the marginal lines the cuticle merges into the general outer layer of the jelly, which sometimes has the appearance of being slightly modified as if to form a thin cuticle over the jelly mass. Beneath the cuticle of the marginal lines lies a band of very finely punctated material, but beyond these punctations, which are densest close beneath the cuticle, not a trace of structure can be detected. There are no cilia, no ctenophoral plates, and no canals.

The terminal curved and serrated lip of one side of the groove (Fig. 5) has the same structure as the marginal lines.

The Central Canal.—The central tubular cavity, with the aboral canal and certain structures associated with them, are

1 I am indebted to the kindness of Mr. W. Fielder for cutting these sections for me.

the only parts where any cellular structure can be detected. The central tube functions, presumably, as an alimentary canal. Its oral opening is just a simple round hole in the jelly, and from this a flask-shaped tube, gradually increasing in size, extends upwards for about four-fifths of the length of the jelly mass (Al. 1). From the centre of the aboral end a small but very definite canal arises, and from the same point four bands take their origin and run down the walls of the tube, alternating in position with the angles of the jelly mass (Figs. 1 and 2). One band corresponds in position with the deep external groove. (Fig. 4, Gl. 1.) In two of the specimens indications can be detected of a very finely attenuated plate-like structure running across to each of the four faces of the jelly from the modified bands. (Fig. 4., Spt.) One of these plates corresponds in position with the deep groove, but in three specimens of which sections were cut I could not distinguish the faintest trace of any structural differentiation representing them. There is not the slightest indication of any canal system other than the central tube. The relationship of the various structures referred to are represented diagramatically in Figure 4.

In some specimens the bands on the wall of the central cavity do not extend as far as the oral opening, but in others they do. Each has clearly a double structure, probably representing two rows of cells, and they (together with the small aboral canal and two special structures to be described later) stain much more deeply than any other part. In Figure 6 a small part of one is shown, together with the adjacent lining of the tube. The band shows no clearly marked outline of cells, only faint indications of this. The protoplasm is reticulated, and when stained stands out in strong contrast to the homogeneous jelly in contact with which it lies like a ribbon. The cells, if such they be, are much flattened out, and contain numerous rounded and irregularly shaped bodies, which stain deeply and sometimes enclose unstained spherical portions. Between the bands the remaining part of the wall of the tube is lined by a single layer of extremely thin cells (Fig. 6, Gl. 2) with nuclei that are smaller than the dark bodies in the bands, and take stain much less deeply. These cells are evidently easily displaced, as in some cases they are absent, and the wall of the tube is actually

formed of jelly. In transverse section they are like the thinnest of pavement cells,

The aboral canal, though small in diameter, is very clearly defined, and as already described, opens into the groove that furrows one side of the organism, and ends in the conical projections on the aboral surface (Fig. 2). The wall of the canal is formed of a definite gelatinous material which stains somewhat more deeply than the ordinary jelly mass, and contains nuclei.

Aboral Organ.—In two specimens there is a curious but definite bunch of projections associated with the aboral canal, lying in the groove close to its external opening. The bunch of processes is transversed by short canals which open into the aboral canal. (Figs. 7 and 8, Al. 2, Al. 3.) The body of the whole mass is made of the same gelatinous material as the main organism, with, however, a tendency to a fibrous formation. Nuclei are scattered irregularly through it, but in addition to these large numbers are arranged in definite relationship to the external surface, and the walls of the canals (Fig. 7), indicating a cellular formation, though no trace of cell outline can be distinguished. Most of the projections are club-shaped, and when stained and cut in section show the structure represented in Figure 8, Pr. There are a number of very definite dark bodies often arranged in two roughly concentric series. The processes are apparently only solid masses of jelly in which these block-like structures are embedded, and their arrangement and general resemblance to what are evidently nuclei in other parts of the processes and in the walls of the canals traversing the latter, suggest the idea that they also are nuclear. If this be so they are of large size in comparison with the cells with which they are associated. It is just possible that some of the canals open on the surface of this enigmatic organ. In three parts there are distinct indications of such openings, but I have not been able to determine the point with certainty.

The Oral Organ.—The only structure remaining to be described is one that is present in four out of the fourteen specimens; no trace of it is to be found in any of the others. In two of the four it is well marked. In Figure 3 a small process can be seen projecting from the oral opening. It is attached to the oral end of one of the cellular bands on the wall of the

central cavity. In the specimen drawn in Figure 3, the process is horse-shoe shaped, which is probably its normal form. It is very small—its relative size can be judged from the figure referred to, which is five times the size of the original. The one represented in Figure 9 is a part of the structure in another specimen that was stained and cut by the freezing microtome. In this the close part of the horse-shoe extended round one quarter of the oral opening, that is, each of the two limbs corresponded in position to one of the cellular bands on the wall of the central cavity. Unfortunately, the section was somewhat broken, and I am unable, amongst the broken parts, to determine definitely the nature of one of the limbs, which appears to be somewhat smaller than the one figured, which has retained its original position. In one of the four specimens in which this organ can be seen, the part attached to the oral surface is alone present, the two limbs of the horse-shoe either not having been developed, or, more likely still, they have been knocked off by the buffeting of the waves on the shore.

The general structure is seen in Figure 9, which represents, as the section was a thick one, a drawing of the solid object. The part attached to the oral surface has the appearance of a semifibrous gelatinous band containing many nuclei, often arranged in rows (d). It is apparently attached along its whole length, but in parts there are remarkable lines of nuclei (e) associated with structures that look like special attachments to the jelly around the mouth. When examining this section under the dissecting miscroscope, I separated the greater part of the horseshoe lying between the two limbs from the oral margin with comparative ease, but at each of the points from which the limbs depend the attachment is a very firm one. It will be noticed that the nuclei, if such they be, of the bands with which this horseshoe-shaped organ is connected are much larger than those of the latter.

The limb shows three different parts, first a fibrous part (a), second a gelatinous part with regularly arranged nuclei (b), and third, a well-marked row of densely packed nuclei (c), lying along a well-marked track between the first and second. At the spot marked N, the limb has a decidedly narrow neck where it is attached to the edge of the oral opening (o.m.). In this region

there are rounded lumps of gelatinous material containing irregularly arranged nuclei. A more or less definite line of nuclei (d) appears to run along the closed end of the horseshoe, and then on to the limb. The general appearance gives the impression that a proliferation of cells is taking place in the attached portion, and in the neck region just alluded to, and that the cells thus formed are wandering on to the limb.. If this be the case, it is possible that the organ is of the nature of a stolon.

General Remarks.—It appears to be impossible to indicate the relationships of this curious organism. The marginal bands at first call to mind those of a ctenophoran, and the main central and aboral canals in like manner suggest the stomodæal and infundibular canals respectively, but there is no trace whatever of any structures resembling the canals that arise from the infundibulum: there is no sense organ: no nerve system: no contractile tissue; the marginal bands are entirely deviod of cilia, and if the deep groove has any relation to a tentacle sheath it is remarkable for its asymmetrical development. The total absence of cells on the external surface is an extraordinary feature, nor do the specimens convey the idea of this being due to accident or bad state of preservation, as they are all in precisely the same condition, and, moreover, the cells of the bands on the canal wall and of the oral and aboral organs are all intact.

Comparison with the members of any other group of animals appears impossible, and the only hypothesis as to its significance that I can offer is one that is suggested by the oral organ. In connection with this I have described above what has the appearance of a proliferation of cells taking place on and close to the part which is attached to the oral margin. From this part, also, it looks very much as if the cells were passing off on to the limb of the organ. If this be so, then it is possible that the latter is an early stage in the development of a stolon, and that this enigmatical organism is a nurse form in the life history of some animal—but as to what this animal may be we have at present not the slightest clue.

EXPLANATION OF PLATES XXIV., XXV.

Fig. 1.—General view from the side. The lateral groove is represented between the marginal bands 1 and 2. Marginal band 4 is seen through the transparent jelly.

Fig. 2.—A portion of the aboral surface on a larger scale, to show the conical elevation cut through by the groove, the upper part of the main central canal with its four bands of cells, the small aboral canal and the processes forming the aboral organ close to the opening of the canal into the groove.

Fig. 3.—Oral view. The mouth is seen in the centre leading directly into the central canal. The numbers 1, 2, 3, 4, correspond in position to the marginal bands seen from the side in Figure 1. The downward growth, conical in form, of the angle associated with marginal band 1, produces the appearance of asymmetry which is probably also accentuated by irregular contraction during preservation.

Fig. 4.—Diagrammatic transverse section across the middle of the body. The marginal bands are numbered, as in the other figures. The relative positions of the marginal bands, lateral groove, ribbons of cells on the walls of the central cavity, and the faintly marked septa (*?*) are shown. The latter are indistinguishable in sections, and only visible in a few specimens.

Fig. 5.—Downward conical process of one angle of the oral, and the oral end of one lip of the lateral groove drawn on a larger scale to show the serrations.

Fig. 5a.—Portion of a marginal band to show the "cuticle" and punctated material beneath. (Zeiss F. Oc. 2.)

Fig. 6.—Part of one of the four ribbon-like bands of cells on the wall of the central cavity, together with the surrounding cells lining the rest of the cavity. All are extremely thin, the latter having much more definite outline than the former. (Zeiss F. Oc. 2.)

Figs. 7 and 8.—Sections across the aboral organ, showing the canals that traverse it, and the regular arrangement of the nuclei in the jelly of which it is formed.

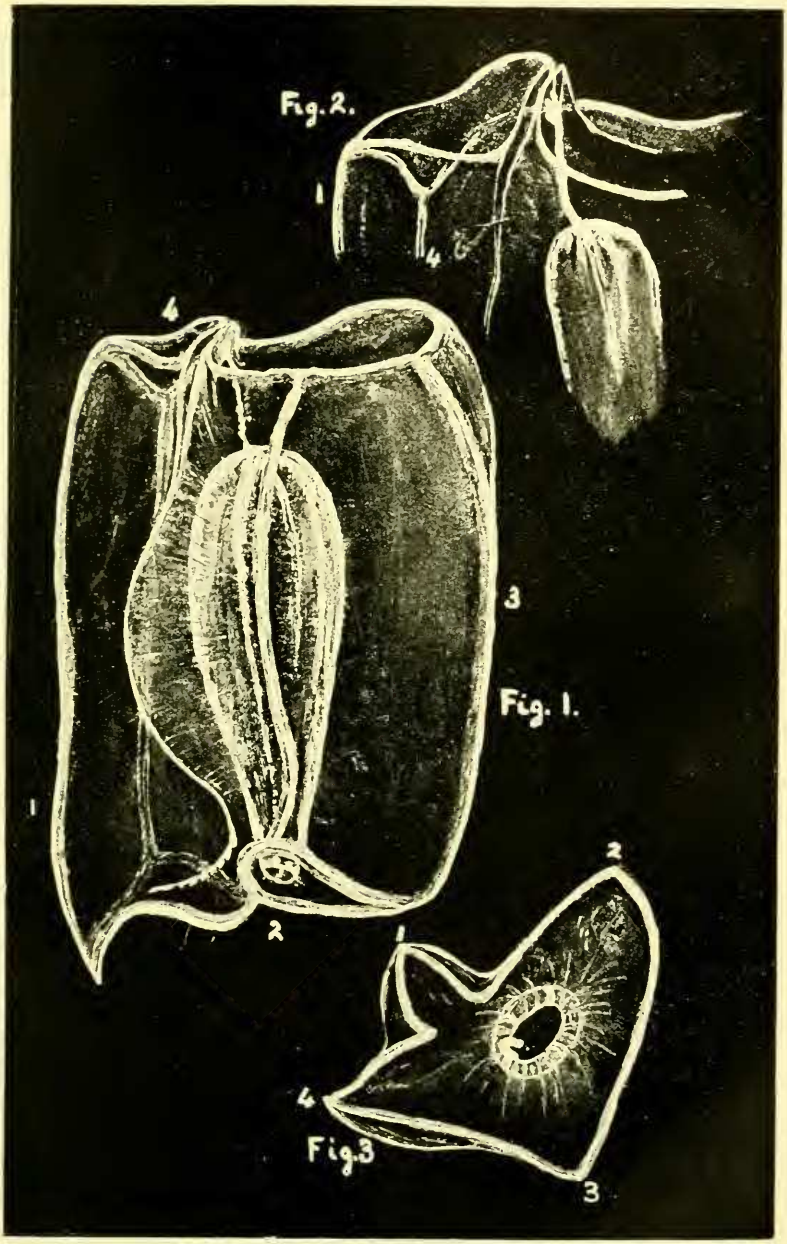
Fig. 2.
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Fig. 1.
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Fig. 3
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